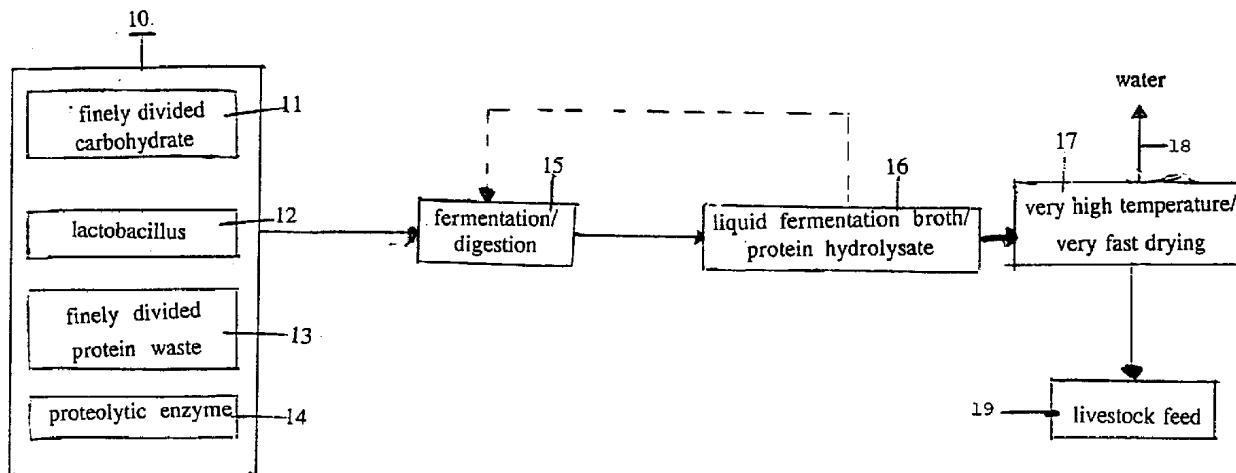


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**(54) PROCEDE POUR CONVERTIR DES SUBSTANCES  
PROTEINIQUES RESIDUELLES EN NOURRITURE  
EQUILIBREE POUR ANIMAUX**  
**(54) PROCESS FOR CONVERTING WASTE PROTEIN SOURCES  
INTO A BALANCED ANIMAL FEED**



(57) Méthode pour préparer de la nourriture, équilibrée, séchée pour animaux. Le processus consiste à faire fermenter simultanément des sources énergétiques de glucides finement divisés, en présence d'un Lactobacillus et à faire digérer les substances protéiniques résiduelles finement divisées en présence d'une enzyme protéolytique. Le produit obtenu est un hydrolysat protéinique/bouillon de fermentation liquide. Celui-ci est séché pendant un temps très court à une température très élevée pour former une poudre. On obtient ainsi le produit alimentaire séché, équilibré pour animaux.

(57) A process is provided for preparing a balanced, dried animal feed. The process comprises simultaneously fermenting finely-divided carbohydrate energy sources in the presence of a Lactobacillus and digesting finely-divided protein waste material in the presence of a proteolytic enzyme. The product obtained is a liquid fermentation broth/protein hydrolysate. Such liquid fermentation broth/protein hydrolysate is dried to a powder at a very high temperature for a very short period. This produces the balanced dried animal food product.

ABSTRACT OF THE DISCLOSURE

A process is provided for preparing a balanced, dried animal feed. The process comprises simultaneously fermenting finely-divided carbohydrate energy sources in the presence of a Lactobacillus and digesting finely-divided protein waste material in the presence of a proteolytic enzyme. The product obtained is a liquid fermentation broth/protein hydrolysate. Such liquid fermentation broth/protein hydrolyzate is dried to a powder at a very high temperature for a very short period. This produces the balanced dried animal food product.

(a) TITLE OF THE INVENTION

Process for converting Waste Protein Sources Into A Balanced Animal Feed

(b) TECHNICAL FIELD TO WHICH THE INVENTION RELATES

5 This invention relates to a process for converting waste protein sources into a balanced animal feed.

(c) BACKGROUND ART

It is known in the art to produce a number of animal feeds as a supplement to,  
10 or even as a substitute for, natural and unprocessed animal fodder. However, these products are either extremely expensive, have only a limited nutritive value, or are particularly poor in essential vitamins.

Attempts have, therefore, been made to use marine products as an animal feed,  
because such marine products not only are abundant and inexpensive, but also have a  
15 particularly high degree of nutritive and generally wholesome value, especially in regard to their contents of amino acids and vitamins.

Some of such marine products used, for instance, in mixtures with comminuted straw or potato-mash, or similar carbohydrate-containing products which are fermented at slightly elevated temperatures, have been mainly so-called "fish offals" comprising the  
20 heads, the spines and similar fishbones, ventricular portions, the skins and sometimes the intestines of fish.

It is also known to treat such marine products, e.g., fish offals, with flour mill by-products, e.g., bran, chaff, residual flour or the like by fermentation with suitable microorganisms to obtain an animal feed.

The known processes utilizing these raw materials suffer from a major drawback  
5 of requiring the handling of large volumes of liquid. Furthermore, most of the known animal feeds obtained from these marine products have a particularly-pronounced taste and odour which is generally repulsive to animals and hence decreases their appetite. Animals fed with such animal feeds, therefore, are prone to consume less food than is desirable and necessary in order to fatten such animals for marketing and slaughtering.  
10 Alternatively, they soon refuse to consume such feed at all. In addition, the meat of animals fed with such animal feeds tend to assume a fishy flavour which makes this meat less well tasting than is generally exacted by the modern consumer.

It is also well-known that several attempts have been made to convert animal protein waste into animal feed, in systems which include aerobic chambers which require  
15 the introduction of oxygen and/or air and which also requires the introduction of cultures. Many such anaerobic processes have been used for the stabilization of municipal sewage sludge, so that such fermentation is for the destruction of the waste matter, rather than the growth of the nutrients in the waste.

In the patent literature, United States Patent 1,604,374, patented October 26,  
20 1926, by N. Albretsson provided a process for the preparation of food for cattle, swine, fowls etc. in which fish or fish-refuse in finely disintegrated condition was intermixed with disintegrated straw fodder, and was then subjected to fermentation.

In addition, potato-mash and, under certain circumstances, a minor quantity of molasses or other binding or flavouring substances or special nutrimental preparations, were included in the mixture. This mixture was allowed to ferment by maintaining a temperature suitable for the fermentation. Subsequent to the fermentation, the mixture  
5 was conventionally dried.

United States Patent 2,806,790, patented September 17, 1952, by R.H. Bedford, provided for the hydrolysis of fish materials, namely, the enzymatic production, from viscera, offal or whole fish and mixtures thereof, of the water soluble proteins and their constituent compounds, hydrolytic products of the nucleic acids, and water soluble  
10 vitamins. The process involved the steps of mixing water, raw tuna viscera, and fish materials together. A minor proportion of sodium chloride, potassium chloride or calcium chloride and urea thiourea or guanidine was added to the mixture. The mixture was heated for a relatively short period of time while maintaining the pH thereof at an acid pH of between 5.5 and 6.0. The heating was continued while maintaining the pH  
15 at an alkaline pH of between 7.8 and 8.0 for a sufficient length of time to hydrolyze the viscera and fish materials. The process also included the separation of these products from the bone and oil and other non-hydrolyzable solid material. The resulting hydrolysate contained 10% to 30% total solids, and remained free flowing when concentrated to 50% to 65% total solids.

20 United States Patent 2,986,469, patented May 30, 1961, by T.M. Krüss, provided a procedure for making animal feed by using de-oiled, concentrated fish solubles. These were admixed with flour mill by-products under specific conditions and were then

subjected to the fermenting action of aerobic, and anaerobic bacteria which can grow in the resulting medium, e.g., Streptococcus cremonis, Lactobacillus leichmannii, or Acetobacter.

United States Patent 3,838,199, patented September 24, 1974, by W.B. Coe et al, the patentee provided, in a laboratory, a feed ingredient having a high crude protein and amino acid content with a high degree of digestibility of the nutrients. By the utilization of anaerobic fermentation and controlling the pressure, temperature, waste concentration and residence time, that feed ingredient was said to be free from undesirable or harmful materials, e.g., estrogenics, antibiotics, and pathogenic organisms.

United States Patent 4,041,181, patented August 9, 1977, by I.E. Burrows et al, provided a solid food product comprising fermented and autolyzed proteinaceous material, usually meat, fish or other animal tissue, bound into coherent pieces by a gelled or coagulated binder and stabilized against microbiological activity by an acid pH value, preferably below 5.5. The starting material was comminuted, and then an acid-producing fermentation culture, for example, of Lactobacillus species, was added with fermentable carbohydrate, and finally the mixture was incubated until the pH value had fallen to 4 or below. A coagulable or gellable binder, e.g., gluten or xanthan gum, was then incorporated, preferably also with antimycotic, and the product was shaped, for example, by extrusion, and was then cooked.

United States Patent 4,288,458, patented September 8, 1981, by E. Barnes, provided a process for producing liquified fish protein by liquifying fish mince using

preformed fish silage as the liquid medium. The unground fish solids were admixed with liquified fish silage in a ratio in the range of from 10:1 to 1:10 by weight and an additive present in at least an amount capable of initiating and accelerating liquefaction of the fish solids, the additive being organic acids, mineral acids, salts thereof, or enzymes.

5 Liquefaction of the fish solids was effected in the presence of the liquified fish silage and at a temperature of at least 20°C.

United States Patent 4,405,649, patented September 20, 1983, by G.A. Jeffreys et al, provided a process for producing a protein fish meal from whole fish, including trash fish, either on board ship or on shore, by using an appropriate proteolytic enzyme 10 which liquified the fish in a digester equipped with high speed blenders. The fish was liquified by heating in the digester. The liquified fish was pasteurized so that the liquid can be stored under refrigeration for several weeks without spoilage. The bones were screened out. Part of the oil was centrifuged out, as necessary. Then, the fish meal was dried. The process is carried out at a pH of 4.5 to 8.0 and a temperature of 30 to 98°C.

15 United States Patent 4,452,888, patented January 5, 1984, by K.I. Yamazaki, provided a low-molecular weight peptide composition mainly based on dipeptides and tripeptides, which was produced by dispersing raw material from any suitable source in water at a concentration of 5 to 20 w/v%. The pH of the dispersion was adjusted to an acid pH of 1 to 4 with an acid. At least two acid proteases were added to the dispersion, 20 and enzymatic proteolysis was allowed to take place for a desired period of time, e.g., 8 to 72 hours, at a suitable temperature, e.g., 25 to 60°C, while suppressing the formation of free amino acids.

United States 4,473,589 patented September 25, 1984 by L.D. Freeman et al provided a process wherein sources of protein, e.g., residues and waste products from processing fish, poultry, pork and beef, as well as single cell microorganisms, were hydrolysed to provide liquid products containing substantially all of the component amino acids, lipids and phosphorus in metabolically useful form. The process involved a brief alkaline treatment by heating (at 120°F to 170°F) at an alkali pH (12 or above) which facilitated liquefaction and enhanced susceptibility to subsequent enzyme hydrolysis with bacterial proteinase at elevated temperatures (100°F to 140°F). Cell rupture and protein denaturation occurred during alkaline treatment and this permitted and facilitated the enzyme rapidly to break down the intact proteins to smaller, more soluble molecules.

United States Patent 4,486,451, patented December 4, 1984 by J.H. Linton et al, provided an unfermented protein animal feed supplement by admixing wet corn bran having an abnormally high moisture content with concentrated corn steep to form a moist cohesive but friable mass, the process eliminating a significant proportion of the energy used in, and pollution occasioned by, prior art process. Alternatively, the above unfermented product may be subjected to a natural fermentation stage using a Lactobacillus microorganism, resulting in a fermented supplement which is microbiologically and gravitationally stable for an extended period of time.

United States Patent 4,528,199, patented July 9, 1987, by N.J. Moon et al, provided a method for the production of silage from a fermentable forage substrate by admixing Lactobacillus plantarum 2B bacteria with a fermentable forage substrate. The bacteria was added in an amount which was effective to lower the pH of the forage

substrate to a pH at which the fermentable forage was stabilized, e.g., to 4.0, and was rendered substantially free of butyric-acid producing bacteria. The fermentation proceeded under anaerobic conditions, and at a temperature of 15 to 45°C.

United States Patent 4,728,517, patented Mach 1, 1988 by W.M. Markham,  
5 provided a process for producing an animal feed, comparable to feathermeal in properties and appearance, from two waste products: namely float sludge and activated sludge. The process used a bulking agent for extracting excess fat from the sludges, while a mixture of the sludges and the bulking agent was being stirred, heated, and dried. Blood, chicken litter, meat scrap, could also provide protein. The bulking agent was, e.g.,  
10 wood chips, wood, wood shavings, shredded cardboard, shredded newspaper, and paunch manure. A primary float sludge was formed from a food wastewater containing protein and fats by admixing air under pressure plus coagulation and flocculation chemicals. The remaining wastewater was treated to form a biological sludge. The biological sludge was mixed with the float sludge. The mixed sludges were aerated and cooked while extracting a sufficient portion of the fat therefrom with a bulking agent. The cooked and partially de-fatted sludge was dried to form a dried mixture having a coating on the  
15 bulking agent. The dried coating was separated from the bulking agent to form a meal. A suitable drying means was a steam cooker of the type usually used for rendering of meat scrap or feather meal. Reaching a moisture content of 4 to 10% usually required  
20 5 to 6 hours.

U.S. Patent 4,759,933, patented July 26, 1988, by Y. Uchida et al, provided protein food products or protein food materials in paste state by grinding fish meat and

treating the fish meat with proteolytic enzymes and/or protein digesting micro-organisms during and/or after grinding to change the properties of the protein contained in the fish meat to reduce or lose the gel forming ability of the protein contained in the fish meat. Food products in solid or spread were prepared by mixing animal and/or vegetable fats and/or oil sources and melting agents to the protein for materials in paste state, agitating.

5 The resulting mixture was agitated while heating to 50° to 100°C. to form a homogeneous mixture. The mixture was then cooled.

United States Patent 4,800,093, patented January 24, 1989, by W.C. Hogan, provided a high moisture animal food product containing meat in which a portion of the meat was replaced with a filamentous fungal biomass, which was produced from the fermentation of a medium, e.g., soybean whey, by a fungi, e.g., Aspergillus oryzae, at a pH of 3.8 to 5.8 and a temperature of 28° to 32°C.

United States Patent 4,820,529, patented April 11, 1989, by Y. Uchida et al, provided a process for preparing a pasty proteinous material or a proteinous food in which crustaceans were boiled and milled under sufficient conditions for inactivating enzymes contained therein, and then proteolytic enzyme(s) and/or micro-organism(s) are allowed to act thereon. Thus, various protein sources, including crustacean meat, remaining in the trunks and carapaces of crustaceans which have been disposed hitherto, can be efficiently utilized.

20 United States Patent 4,861,602, patented August 29, 1989 by Y. Uchida et al, provided a process whereby fish bodies, from which the internals and/or skins were optionally removed, were treated with a protease to give a gain in soluble nitrogen con-

tained in the treated material, based on the total nitrogen contained in the starting material of 3 to 50%, thereby to give a slurry. Fish bones, fish oil, partially decomposed fish proteins and an aqueous solution containing water-soluble components were separated and recovered from the slurry.

5                 United States Patent 4,842,871, patented June 27, 1989, by J.E. Hill, provided a method of preserving agricultural products comprising treating these products with an effective amount of Lactobacillus plantarum ATCC 53187, or mutants thereof, and the treating organism.

United States Patent 4,940,662, patented July 10, 1990, by K.I. Yamazaki et al,  
10 provided a method of producing a low-molecular weight peptide mixture. The process include the step of dissolving a first protease in a buffer solution adjusted to an optimum pH for the protease, ranging from a pH of 3 to 10. At least one first protein in a buffer solution having a pH of from 3 to 10 and a concentration of 10 to 60% by weight of the protein material was added, and was thoroughly mixed in the solution. A solution of an  
15 ester of at least one amino acid formed by esterification of the amino acid with an alcohol was then added, the pH of such buffer solution being optimum for the incorporation of the amino acid in the starting protein in the presence of the first protease in a plastein reaction. The ester of at least one amino acid was reacted with the starting protein in a plastein reaction in the mixed solution, whereby at least one amino acid was  
20 covalently incorporated into the starting protein to produce a plastein reaction solution containing a modified protein. The modified protein was hydrolyzed using at least one second protease having a different specificity from at least one first protease to produce

a low-molecular weight peptide mixture having an amino acid content which had different proportions of amino acids than did the starting protein. The low-molecular weight peptide mixture comprised a major proportion of dipeptides and tripeptides and contained not more than 15% by weight of free amino acids and not more than 20% by weight of 5 a high-molecular weight fraction of compounds having a molecular weight of not lower than 700 and which was removable by gel filtration, was separated from the solution.

United States Patent 4,963,370, patented October 16, 1990, by Y. Uchida et al, provided a process for producing a proteinous material, which comprised coarsely grinding fish bodies including bones and/or shells, from which the internals had been 10 removed, optionally together with head and/or skins, and defatting those coarsely ground fish bodies if required. Then, the process involved the step of: either fermenting the coarsely ground fish bodies with an enzyme and/or a microorganism, then inactivating the enzyme and/or microorganism and then finely grinding the fermented material; or finely grinding the coarsely ground fish bodies, then fermenting them with an enzyme 15 and/or a microorganism, and then inactivating the enzyme and/or microorganism; or finely grinding the coarsely ground fish bodies while fermenting them with an enzyme and/or a microorganism, and then inactivating the enzyme and/or microorganism.

United States Patent 4,976,973, patented December 11, 1990, by Y. Shirakawa et al, provided a process for producing a protein-rich fish meal and a fish oil, which 20 comprised treating fish bodies with a protease acting at a relatively low temperature to give a slurry and dividing and drying the slurry at a relatively low temperature. The

products thus obtained scarcely underwent thermal denaturation and contained a large amount of partially decomposed protein.

United States Patent 5,053,233, patented October 1, 1991, by J.J. Setälää et al provided a process of preserving forage. The bacterial strain Lactobacillus plantarum 5 was preferably used in combination with at least one other preservative, e.g., cellulase and organic acid, and/or in combination with at least one other lactic acid bacterium.

United States Patent 5,518,741, patented May 21, 1996, by G.S. Choudhury, provided a method of making a food product from fish muscle. The fish were, e.g., arrowtooth flounder, Peruvian hake, Pacific whiting, Yellowfin sole, or menhaden, 10 containing protease enzyme. The method involved distributing the enzyme substantially throughout the fish muscle. The fish muscle was autolyzed at a temperature which was sufficient to permit protease degradation therein During or after the autolyzing, the fish muscle was dried at a temperature which was sufficient to reduce moisture content and thus to form a dried autolyzed fish muscle. The dried fish muscle was then reduced to 15 powder form. The powder was mixed with a starchy and/or proteinaceous material to form a mixture. That mixture was introduced into an extruder, and the mixture was then extruded in an elevated temperature extrusion process.

United States Patent 5,529,793, patented June 26, 1996, by B.E. Garner et al, provided a composition for improving the utilization of feedstuffs by ruminants through 20 the introduction of the composition into the rumen of the ruminants. The composition included a mixture of a lactic acid producing bacteria culture and a lactate-utilizing bacteria culture. The bacteria cultures were admixed with an animal feedlot diet, e.g.,

selected corn, dried grain, alfalfa, or corn meal. The lactic acid-producing bacteria culture produced lactic acid in the rumen of the ruminant in order to promote the growth of lactate-utilizing bacteria culture, thereby to prepare the rumen for a feed diet which generates high levels of lactic acid in the rumen. The lactate utilized the bacteria culture consuming lactic acid produced in the rumen, thereby decreasing levels of lactic acid in the rumen. Examples of lactic acid-producing bacteria included the following:

5                   Lactobacillus acidophilus; Lactobacillus plantarum; Lactobacillus casei; Lactobacillus lactis; Lactobacillus enterii; Lactobacillus fermentum; Lactobacillus delbruckii; Lactobacillus helveticus; Lactobacillus curvatus; Lactobacillus brevis; Lactobacillus bulgaricus; and Lactobacillus cellobiosuus.

10                  United States Patent 5,534,271, patented July 9, 1996, by D.R. Ware et al, provided a process for improving the utilization of feedstuffs by ruminants, especially during the transition from a roughage diet to a feedlot diet. The process comprised mixing a lactic acid-producing bacteria culture and a lactate utilizing bacteria culture. These cultures were admixed with a ruminant feedlot diet essentially consisting of corn, dried grain, alfalfa, and corn meal to form a composition. That composition was administered orally to the ruminants.

15

**(d) DESCRIPTION OF THE INVENTION**

20                  These patented procedures have not been generally commercially successful. Accordingly, it is an object of one aspect of the present invention to provide animal feed

from waste protein sources, the animal feed being inexpensive and being easily produced in large quantities.

It is an object of another aspect of the present invention to provide such animal feed which is rich in amino acids and vitamins.

5 An object of yet another aspect of this invention to provide a process for producing such animal feed.

By one broad aspect of this invention, a process is provided for preparing a dried animal food product, comprising: substantially-simultaneously fermenting finely divided carbohydrate energy sources in the presence of a Lactobacillus and digesting 10 finely divided protein waste material in the presence of a proteolytic enzyme, thereby to obtain a liquid fermentation broth/protein hydrolysate; and subjecting that liquid fermentation broth/protein hydrolysate to a drying process at a very high temperature for a very short period, thereby to produce the dried animal food product.

By one variant of this aspect of the invention, the finely-divided carbohydrate 15 energy source includes a minced forage crop. By one variation of that variant, the minced forage crop is timothy, alfalfa, clover, bluegrass, reedtop, marine grasses, velvet beam, rye, barley, or oats.

By another variant of this aspect of the invention, the finely-divided carbohydrate 20 energy source includes finely divided tuber. By one variation of that variant, the tuber is potatoes, cassava, Jerusalem artichokes, sweet potatoes, yams, arrowroot, colocasia, turnips, or sugar beets.

By yet another variant of this aspect of the invention, the carbohydrate energy source includes a sugar. By one variation of that variant, the sugar is sucrose or molasses.

By still another variant of this aspect of the invention, the finely-divided protein waste material may be one or more of the following: fish wastes, including whole trash fish, fish left after filleting, fish solubles, fish intestines, and any other material which is a byproduct of the fishing industry and fish processing; poultry wastes of all types, including blood, internal organs, feathers, beaks, heads, feet, whole birds and eggs; pork skins or other pork residues, including all internal organs, containing protein and fats, or any pork tissues or byproducts, and beef tissues, beef intestines, or other waste products, including all internal organs, derived from beef manufacture containing proteins and fats and animal blood.

By still another variant of this aspect of the invention, the Lactobacillus is L. plantarum, L. leichmannii, L. delbrueckii or L. bulgaricas.

By a still further variant of this aspect of the invention, the proteolytic enzyme is a mammalian serine protease, a bacterial serine protease, an enzyme derived from Streptococcus plantarum, or trypsin.

By a further variant of this aspect of the invention, the temperature of fermentation/hydrolysis takes place at 27 to 39°C, at a pH of less than 7 for a time within two days.

By yet a further variant of this aspect of the invention, the drying takes place at a temperature of at least 2000°C for a time of less than 1 sec. By one variation of that

variant, such drying steps are sufficient to dry the liquid fermentation broth/protein hydrolysate to a solid having a moisture content of 10 to 20%. By another variation of that variant, the dried solid is subjected to the step of pelletizing or extruding.

In carrying out the process of an aspect of the present invention, the finely-divided protein material may be viscera, offal or whole fish which may consist of the following: raw tuna viscera, which may consist of the whole of the alimentary canal, the pancreas, the ceca, and includes the liver and all the organs of the body cavity. Fresh or frozen fresh whole fish or offal, which consists of heads, tails and adhering muscle, the viscera, and damaged whole fish not used for human consumption; and heated whole fish or offal, which consists of fresh or frozen whole fish or offal which has been heated.

Other examples of fish meat which is usable in the process of an aspect of this invention ("fish" herein includes marine animals usable for ordinary fish food processing) includes fish meat collected from various fishes, fish meat "Surimi", "Otoshimi", fresh or frozen, and various kind of fish meat, whether processed or non-processed.

Other examples of fish include pollock, cod, flatfish, turbot, perches, sardines, mackerels, pikes, croakers, horse mackerel, squids, tuna, skipjacks, swordfish, yellow tail, salmon, trout, herring, shark, octopus, shrimps, whales, scabbard fish, bastard halibut, warazuka, herring, saury, round herring, Alaska pollack, anchovy, pillhard, saurel, bonito, buna salmon, cuttlefish, and shell fishes. Examples of shell fishes include those crustaceans which are conventionally employed in processing fishery products, e.g., crabs, king crab, latrpillia phalangium, ternner crab, red turnner crab, paralithodes breripes, maja spirigera, mask crab, leucosia obtasifrons, hairy crab and blue crab and

shrimps, e.g., tiger prawn, *Metapenaeus joyneri*, krill, *Palaemon nipponensis*, shrimp, lobster, crawfish and *Upogekia major*.

The proteinous material which may be used in a process of an aspect of the present invention may comprise animal protein sources, vegetable protein sources, animal and vegetable fat sources, and carbohydrate sources. For example, the process of an aspect of the present invention may further use, as typical finely-divided protein waste materials, the following: animal blood; poultry wastes of all types, including blood, internal organs, feathers, beaks, heads, and feet, whole birds, and eggs; pork skins or other pork residues, including all internal organs containing protein and fats, or any pork tissues or byproducts or organs; beef tissues, e.g., beef intestines, or other waste products, including all internal organs, derived from beef manufacture containing proteins and fats.

Examples of animal protein food source which may be used in a process of an aspect of the present invention may include protein materials, for example, milk and milk products, skim milk, condensed milk, whole milk powder, skim milk powder, modified milk powder, butter, cheeses, and cream; livestock meat, e.g., beef, pork, mutton, and chicken; processed livestock meat, e.g., smoked meat, and dried meat; egg and egg products, e.g., whole eggs, frozen eggs, dehydrated eggs, egg white, and egg-yolk; and other animal protein sources, e.g., liver.

The finely-divided carbohydrate energy food source which may be used in a process of the aspect of the present invention may include vegetable protein materials, for example, soy bean, peanut, cotton seed, sesame, sunflower, wheat, and their oil

extract products and concentrated protein desired therefore and isolated proteins and the like.

Other examples of such finely-divided carbohydrate sources include agricultural products containing a large amount of carbohydrates, e.g., rice, wheat, corn, potato and sweet potato; powdery products obtained therefrom; starch products obtained therefrom, e.g., rice starch, wheat starch, corn starch and potato starch; processed or denatured starch products, e.g.,  $\alpha$ -starch and dextrin; sugars, e.g., sucrose, honey and starch sugar; and fresh apple, orange, strawberry and grape, or juices therefrom.

Examples of proteolytic enzymes which may be used in a process of an aspect of the present invention may include: proteinases, e.g., acrosin, urokinase, uropepsin, elastase, enteropeptidase, cathepsin, kallikrein, kinase 2, chymotrypsin, chymopapain, collagenase, streptococcus peptidase-A, subtilisin, thermolisin, trypsin, thrombin, papain, pancreatopeptidase, ficin, plasmin, renin, reptilase, and rennin, aminopeptidases, e.g., arginin aminopeptidase, oxidase, leucine aminopeptidase, angiotensinase, angiotensine converting enzyme, and insurinase; carboxy peptidase, e.g., arginine carboxy peptidase, kinase-1, and chroidpeptidase; peptidases, e.g., carnosinase, prolidase and similar peptidase and other protein digesting enzymes and their variants and prosthetic group.

The mixture may be finely ground by using a grinder, e.g., a stone mill in such a manner as to give a particle size of a proteinous material, in particular bones and shells, of  $200\text{ }\mu$  or less, preferably  $100\text{ }\mu$  or less.

This resulting low-moisture product has potential as a feed for livestock (including pigs, chickens, cows), as well as a feed for use in aquaculture, and as a feed for fur-bearing animals.

5

(e) DESCRIPTION OF THE FIGURES

In the accompanying drawings,

Figure 1 is a schematic representation of a broad generic aspect of the basic process of an embodiment of this invention;

10

Figure 2 is a schematic representation of one specific aspect of the basic process of another embodiment of this invention;

Figure 3 is a schematic representation of another specific aspect of the basic process of yet another embodiment of this invention; and

15

Figure 4 is a flow chart, in schematic form, of a series of apparatus units which may be used in carrying out the process of a broad generic aspect of an embodiment of this invention.

(f) ONE MODE FOR CARRYING OUT THE INVENTION

(i) Description of Figure 1

As seen in Figure 1, a blend of finely divided carbohydrate material in zone 11  
20 and finely divided protein waste in zone 13 is subjected, along with a suitable Lactobacillus in zone 12 and a suitable proteolytic enzyme in zone 14 to a substantially-simultaneous fermentation/digestion in zone 15, the outflow therefrom being liquid

fermentation broth/protein hydrolysate in zone 16. Such liquid fermentation broth/protein hydrolysate is subjected to very high temperature/very fast drying in zone 17. Water is expelled at 18. The dried product is a livestock feed at zone 19.

This process may take place at a temperature of 27 - 39°C at a pH of less than 5 7 for a time up to two days. The drying may be conducted at a temperature of 2000°C or higher for a time of 1 sec or less.

(ii) Description of Figure 2

As seen in Figure 2, minced tubers in zone 202, minced fish, including natural proteolytic enzymes, in zone 204, a source of digestible energy in zone 206 and a 10 selected Lactobacillus in zone 208 are mixed together in zone 210 and are fermented/digested together in zone 212 to provide a liquid lactic acid-containing fermentation broth/fish hydrolysate in zone 214. Such liquid lactic acid-containing fermentation broth/fish hydrolysate is dried very rapidly at a high temperature in zone 216, where water is simultaneously expelled at 217. The dried product is livestock feed 15 at zone 218.

The above described process may be carried out within a temperature range of about 30°C for a time of less than two days. The drying may take place at a temperature of 2000°C for a time of less than 1 sec.

(iii) Description of Figure 3

20 As seen in Figure 3, the minced tubers, e.g., potatoes in zone 303, the minced fish, e.g., cod, which also contains naturally-occurring proteolytic enzymes in zone 304, a forage crop, e.g., hay in zone 306, a source of a sugar, e.g., molasses in zone 308,

and a lactic-acid producing bacteria, e.g., L. plantarum, in zone 312, are mixed together in zone 310. This mixture is then substantially-simultaneously fermented and digested, (as shown by arrow 313), to provide a lactic acid-containing fermentation broth/liquid fish hydrolysate in zone 314. The liquid lactic acid-containing fermentation broth/liquid fish hydrolysate in zone 314 is dehydrated very rapidly at a very high temperature at zone 316, with water being removed at 318. The product is livestock feed at zone 320.

The above-described process may be carried out within a temperature range of 30°C. for a time of less than two days. The drying may take place at a temperature of 2000°C for a time of less than 1 sec.

10 (iv) Description of Figure 4

As seen in Figure 4, the raw products, e.g., those used in zones 10, 210, 310, in Figures 1, 2, or 3 respectively, are ground together in a suitable grinder or hammer-mill 402 to provide a reactant product of slurry consistency.

15 The minced raw materials, in free flowing slurry form, are passed via line 504 to be fermented/digested in reactor 506 (which is equivalent to zone 15 in Figure 1, zone 212 in Figure 2, and zone 314 in Figure 3). The reaction may be carried out at a temperature of about 30°C for a time of less than two days. For more efficient operation, the reaction products are withdrawn via line 508 and are re-cycled by pump 510 and line 512 back to fermenter/digester 506. Alternatively, efficient operation may 20 be achieved by continuous stirring of the contents of the fermenter/digester 506.

These reacted materials are now in the form of a liquid lactic acid-containing fermentation broth/protein hydrolysate and are passed from fermenter/digester 508 via

line 514 to a suitable, very rapid, high temperature, dryer 518. Heat may be recovered in heat exchanger 522 for further use (not shown) by means of cycle lines 524, 526.

The dried product emerging from dryer 518 via line 528 has a moisture content of about 10 to about 20% by weight and passes to a pelletizer/extruder 530. The final pelletized or extruded product is withdrawn via line 532 to product storage 534.

5                   (g) OPERATION OF PREFERRED EMBODIMENTS

This resulting low-moisture product has potential as a feed for livestock (including pigs, chickens, cows), and for fur-bearing animals, as well as feed for use in aquaculture. As will be seen from the examples hereunder, it has been used successfully 10 as a fish feed.

The nutritional value of products of the present invention as a protein supplement for fish feed and other aquaculture uses was evaluated. The nutrient (protein, fat, carbohydrate, mineral and vitamin) and energy content of a feed ingredient can be determined by chemical analysis. However, analytical data represents the total amount 15 of nutrient present in a feed supplement and does not provide information on the digestibility of nutrients nor their availability to fish. Nutrients, e.g., protein, fat, and carbohydrate of a feed ingredient, must be hydrolyzed prior to absorption by the fish. During digestion, protein, fat, and carbohydrate are broken down to produce amino acids, fatty acids, and glucose which supply the "digestible energy" as fuel for 20 metabolism. In order to determine the suitability of a feed ingredient, information on

digestible energy and digestibility co-efficients of protein, fat, and carbohydrate are necessary.

Digestibility measurements are difficult with fish because of the problem of separating feces from water and also avoiding contamination of feces with uneaten food.

5 Several methods have been developed for the collection of feces for fish digestion studies. These include manual stripping of fish, intestinal dissection, feces recovery from aquarium water by netting or rotating screen filters and settling columns. The method used herein involves a settling column.

10 Several products of the present invention have been evaluated as a source of protein and lipid in Atlantic salmon diets in both freshwater and seawater. Products of this invention (referred to as "Feed Stock Base") was incorporated into a standard test diet at 30% and 50% levels. Two different batches of Feed Stock Base were tested. Chromic sesquioxide was used as an insert digestion indicator.

15 **METHOD**

Diets

1. Reference diet
2. Reference diet (70%) + Herring Meal (30%)
3. Reference diet (70%) + Feed Stock Base # 1 (30%)
- 20 4. Reference diet (70%) + Feed Stock Base # 1 (50%)
5. Reference diet (50%) + Feed Stock Base # 2 (30%)
6. Reference diet (50%) + Feed Stock Base # 2 (50%)

Each of the diets was finely ground and steam pelleted. The compositions of the reference diets and of the test diets is shown in Tables 1-3.

Table # 1 Composition of Reference Diet

	<u>Ingredient</u>	<u>Amount (%)</u>
	Herring meal	35.0
	Soybean meal	18.0
	Wheat middlings	26.6
10	Wheat gluten meal	4.0
	Carboxy methyl cellulose	3.0
	Vitamin mix	3.2
	Mineral mix	2.0
15	Choline chloride	0.2
	Herring oil	8.0

Table # 2 Reference and Test Diets for Digestibility Study

	<u>Ingredient</u>	<u>diet #1</u>	<u>diet #2</u>	<u>diet #3</u>	<u>diet #4</u>	<u>diet #5</u>	<u>diet #6</u>
20	Reference diet	100%	69.3%	69.3%	49.3%	69.3%	49.3%
	Herring meal	0%	30%	0%	0%	0%	0%
	Feed Stock Base #1	0%	0%	30%	50%	0%	0%
	Feed Stock Base #2	0%	0%	0%	0%	30%	50%
25	Chromic oxide	0.7%	0.7%	0.7%	0.7%	0.7%	0.7%

Table # 3 Composition of test ingredients

	<u>Test ingredient</u>	<u>Dry matter</u>	<u>Ash</u>	<u>Protein</u>	<u>Lipid</u>	<u>Carbohydrate*</u>
30			<u>%</u>			
	Feed Stock Base 1	86.1±0.04	14.9±0.08	50.8±0.44	2.7±0.11	
35	Feed Stock Base 2	81.1±0.31	11.4±0.21	35.1±1.03	5.2±0.04	27.0±0.59
	Herring Meal	95.3±0.22	12.9±0.01	68.1±0.67	12.0±0.07	

\*Carbohydrate refers to sugar and starch combined

### Fish

40 Atlantic salmon fingerlings were used for freshwater studies. Atlantic salmon smolts which had been already acclimated to seawater for two months were used in this

experiment. Each diet was fed to three groups of fish maintained in individual tanks. The water temperature was maintained between 15 and 16°C throughout the experiment in all the aquaria. The salinity of seawater was 32 ppt. The water temperature was monitored daily. Fish were fed all they would eat 3 times a day. After a ten day adjustment period, feces were collected from each dietary groups for a minimum of 10 days (pooled in two groups).

#### Metabolic Tanks and Feces Collection

The tank system used were three tanks (55 x 50 x 35H cm) set up with a common drain pipe which led into a single outside stand pipe. Three sets of these tanks were used, two of which were filled with fresh water. The remaining set of tanks were filled with salt water. Each of the fresh water tanks were filled with 35 salmon fingerlings with an average weight of 43.9 grams. The saltwater tanks were each filled with 20 Atlantic salmon smolts with an average weight of 96.1 grams. The water flow was controlled to give the least settling of feces in the drainage system and the maximum settling of feces in the collection tube (10 ID x 45H cm) which was connected under the straight bottom of the stand pipe. The feces trapped in the bottom of the collection tube were held in quiet water until the daily morning collection. This minimized leaching of nutrients from the feces.

Three feedings were carried out during the time period 08:30 to 15:30 h. One hour after the final feeding the collection tube was emptied and the tanks and common drain pipe were cleaned to remove any residual feces. At 08:30 the following morning

the feces which accumulated during the night was collected. The feces which settled between 16:00 and 08:30 h were considered free from uneaten feed and representative of feces for the 24 h period. The collected feces were centrifuged at 5000 rpm for twenty minutes and the supernatant liquid was decanted. The feces were then freeze-dried and subsequently analyzed for undigested nutrients and digestion indicator.

#### Reference Diet and Test Ingredients

Feeding a single ingredient or test substance assumes that there are no interactions between classes affecting digestibility. The use of a reference diet and this diet plus 30% and 50% of the test ingredient was designed to simulate practical feeding conditions. The difference in proximate composition between the reference diet and the test diets was assumed to be entirely a function of the test ingredient. The mixed diets were steam pelleted using a pellet size of 1/8 of an inch.

Crude protein ( $N \times 6.25$ ) of diets and feces was determined using A.O.A.C. (1975) method; gross energy was determined by combustion in a Parr adiabatic oxygen bomb calorimeter; crude fat was measured by acid hydrolysis method; starch and sugar, a well-known conventional method, and  $Cr_2O_3$  was determined, using a well-known conventional method.

### Calculation of Results

$$\frac{\% \text{ nutrient in diet}}{\% \text{ Cr}_2\text{O}_3 \text{ in diet}} - \frac{\% \text{ nutrient in feces}}{\% \text{ Cr}_2\text{O}_3 \text{ in feces}}$$

5

$$1. \quad \text{digestibility coeff.} = \frac{\% \text{ nutrient in diet}}{\% \text{ Cr}_2\text{O}_3 \text{ in diet}} \times 100$$

10

$$2. \text{ digestibility coeff.} = \frac{100}{\text{of test ingredient}} (\text{dig. coeff. of } - \frac{70}{30 \text{ test diet}} \text{ dig. coeff. of } ) \frac{100}{100 \text{ reference diet}}$$

15

20

$$4. \quad \frac{\text{digestible energy}}{\text{gram of feed}} = \frac{\text{gross energy}}{\text{gram of diet}} - \frac{\text{feces energy}}{\text{gram of diet}}$$

25

$$5. \frac{\text{digestible energy}}{\text{gram of test ingredient}} = \frac{100}{30} (\text{digest. energy} - \frac{70}{100} (\text{digest. energy}))$$

30

The results are shown in Tables 4, 5, and 6.

35

Table # 4 Digestibility coefficients and digestible energy of test ingredients

40	Diet Number	Test Ingredient	Digestibility (%)			Digestible Energy (Kcal/Kg)
			Protein	Lipid	Energy	
<u>Energy</u>						
45	fresh water # 2	30% Herring Meal	95.6±0.85	95.9±0.90	93.8±1.65	5150
	salt water # 2	30% Herring Meal	93.2±0.56	95.9±1.02	92.7±1.96	5034
	fresh water # 3	30% Feed Stock Base 1	93.7±2.24	92.5±1.82	90.3±2.76	4156

	salt water # 3	30% Feed Stock Base 1	$93.7 \pm 1.65$	$95.9 \pm 2.99$	$89.3 \pm 1.62$	3959
	fresh water # 4	50% Feed Stock Base 1	$93.6 \pm 0.66$	$91.2 \pm 0.29$	$88.7 \pm 0.77$	4078
	salt water # 4	50% Feed Stock Base 1	$91.5 \pm 0.75$	$95.3 \pm 0.95$	$85.4 \pm 1.26$	3809
	fresh water # 5	30% Feed Stock Base 2	$93.4 \pm 0.31$	$95.8 \pm 3.12$	$84.6 \pm 1.06$	3658
5	salt water # 5	30% Feed Stock Base 2	$93.7 \pm 0.23$	$95.9 \pm 0.89$	$82.6 \pm 0.48$	3558
	fresh water # 6	50% Feed Stock Base 2	$92.3 \pm 0.18$	$90.4 \pm 2.11$	$82.8 \pm 1.01$	3727

Table 5 Data on Dry Matter Basis with Salt Water Adjustment

10	Sample	% Protein	% Fat	% Ash	% Cr <sub>2</sub> O <sub>3</sub>	Gross Energy
15	feed # 1	42.628	14.57	8.393	0.7000	5.375
	feed # 2	50.003	12.897	10.019	0.6647	5.2979
	feed # 3	46.038	11.311	12.037	0.6808	5.0345
	feed # 4	48.922	9.37	13.733	0.6755	4.9027
	feed # 5	45.168	14.789	9.863	0.8169	5.0491
	feed # 6	47.304	13.999	10.713	0.8156	4.9295
20						
25	FW#1-1	12.586	6.353	15.578	1.7365	3.8693
	FW#1-2	11.6	5.906	16.509	2.4845	3.8207
	SW#1-1	14.421	7.723	16.044	2.7535	3.5272
	SW#1-2	14.238	8.234	16.044	2.6113	4.0132
30	FW#2-2	14.453	4.212	26.884	3.278	3.332
	FW#2-2	13.802	4.252	28.042	2.9495	3.3281
	SW#2-1	19.356	4.476	27.463	2.9895	3.127
	SW#2-2	18.694	4.014	27.463	2.9691	3.3027
35	FW#3-1	11.906	4.372	29.714	3.009	3.1797
	FW#3-2	12.782	3.682	28.637	2.7795	3.178
	SW#3-1	12.919	4.792	29.176	2.5545	2.9824
	SW#3-2	12.414	3.121	29.176	2.7085	3.0245
40	FW#4-1	14.932	4.0127	33.647	2.846	2.9989
	FW#4-2	13.761	3.8747	35.971	2.804	2.8436
	SW#4-1	12.853	2.448	35.809	2.253	2.6212
	SW#4-2	13.024	2.005	35.809	2.1395	2.6272
45	FW#5-1	12.434	5.643	19.209	3.3786	3.8544
	FW#5-2	12.469	4.367	20.15	3.3223	3.6982
	SW#5-1	12.356	4.462	19.679	3.0076	3.3858
	SW#5-2	11.919	3.74	19.679	2.9405	3.3602
50						

FW#6-1	13.518	6.964	20.63	3.4746	3.7894
FW#6-2	12.272	5.62	21.892	3.2164	3.6412

5

Table # 6 Digestibility coefficients of whole feed

	sample	Digestibility (%)			Digestible Energy
		Protein	Lipid	Energy	
10	FW#1-1	92.447	88.846	81.585	4.3852
	FW#1-2	92.333	88.579	79.972	4.2985
	SW#1-1	91.399	86.524	83.317	4.4783
	SW#1-2	91.046	84.851	79.985	4.2992
15	FW#2-1	94.138	93.377	87.246	4.6222
	FW#2-2	93.78	92.57	85.842	4.5478
	SW#2-1	91.393	92.284	86.876	4.6026
	SW#2-2	91.63	93.039	86.043	4.5585
20	FE#3-1	94.148	91.254	85.71	4.318
	FW#3-2	93.199	92.026	84.538	4.256
	SW#3-1	93.521	88.709	84.211	4.2396
	SW#3-2	93.222	93.064	84.899	4.2742
25	FW#4-1	92.755	89.834	85.481	4.1909
	FW#4-2	93.223	90.037	86.027	4.2177
	SW#4-1	92.122	92.167	83.969	4.1168
	SW#4-2	91.594	93.242	83.0811	4.0732
30	FW#5-1	93.344	90.774	81.685	4.1243
	FW#5-2	93.212	92.739	82.134	4.1470
	SW#5-1	92.57	91.805	82.043	4.1424
	SW#5-2	92.669	92.975	81.841	4.1322
35	FW#6-1	93.292	88.322	82.124	4.4048
	FW#6-2	93.421	89.82	81.408	4.0130
40					
45					

## (h) CLAIMS:

1. A process for preparing a dried animal food product, comprising: substantially-simultaneously fermenting finely divided carbohydrate energy sources in the presence of a Lactobacillus, and digesting finely divided protein waste material in the presence of a proteolytic enzyme, thereby to obtain a liquid fermentation broth/protein hydrolysate; and subjecting said liquid fermentation broth/protein hydrolysate to a drying process at a very high temperature for a very short period of time, thereby to produce said dried animal food product.
2. The process of claim 1 wherein said finely-divided carbohydrate energy source includes a minced forage crop.
3. The process of claim 2 wherein said minced forage crop is selected from the group consisting of timothy, alfalfa, clover, bluegrass, redtop, marine grasses, velvet beam, rye, barley, and oats.
4. The process of any one of claims 1 to 3, inclusive, wherein said finely divided carbohydrate energy source includes finely divided tubers.
5. The process of claim 4 wherein said tuber is selected from the group consisting of potato, cassava, Jerusalem artichoke, sweet potato, yam, arrowroot, colocasia, turnip, and sugar beet.
6. The process of any one of claims 1 to 5, inclusive, wherein said finely-divided carbohydrate energy source includes a sugar.

7. The process of claim 6 wherein said sugar is selected from the group consisting of sucrose and molasses.

8. The process of any one of claims 1 to 7, inclusive, wherein said finely divided protein waste material is selected from the group consisting of fish wastes, whole trash fish, fish left after filleting, fish solubles, fish intestines, and any other material which is a byproduct of the fishing industry and fish processing industry; poultry wastes of all types, including blood, internal organs, feathers, beaks, heads, and feet, whole birds and eggs; pork skins or other pork residues, including all internal organs, containing protein and fats, or any pork tissues or byproducts and beef tissues, beef intestines, or other waste products, including all internal organs, derived from beef manufacture containing proteins and fats and animal blood.

9. The process of any one of claims 1 to 8, inclusive, wherein said Lactobacillus is selected from the group consisting of L. plantarum, L. leichmannii, L. delbrueckii and L. bulgaricas.

10. The process of any one of claims 1 to 9, inclusive, wherein said proteolytic enzyme is selected from the group consisting of a mammalian serine protease, a bacterial serine protease, an enzyme derived from Streptococcus plantarum, and trypsin.

11. The process of any one of claims 1 to 10, inclusive, wherein said protein starting material is cod, pollack, caplin, herring, mackerel, salmon or seal, wherein said Lactobacillus is L. plantarum; and wherein such enzyme is trypsin.

12. The process of any one of claims 1 to 11, inclusive, wherein said temperature of fermentation/digestion takes place at 27 to 39°C at a pH of less than 7 and for a time within two days.
13. The process of any one of claims 1 to 12, inclusive, wherein said drying takes place at a temperature of at least about 2000°C for a time of 1 sec or less.
14. The process of claim 13 wherein said drying is sufficient to dry said liquid fermentation broth/protein hydrolysis to form a dried solid having a moisture content of 10 to 20%.
15. The process of any one of claims 1 to 14, inclusive, wherein said Lactobacillus is L. plantarum; wherein said enzyme is trypsin; wherein said fermentation temperature is 30°C; and wherein said drying takes place at a temperature of at least about 2000°C for a time of less than about 1 sec.
16. The process of any one of claims 1 to 15, inclusive, including the step of pelletizing or extruding said dried solid.

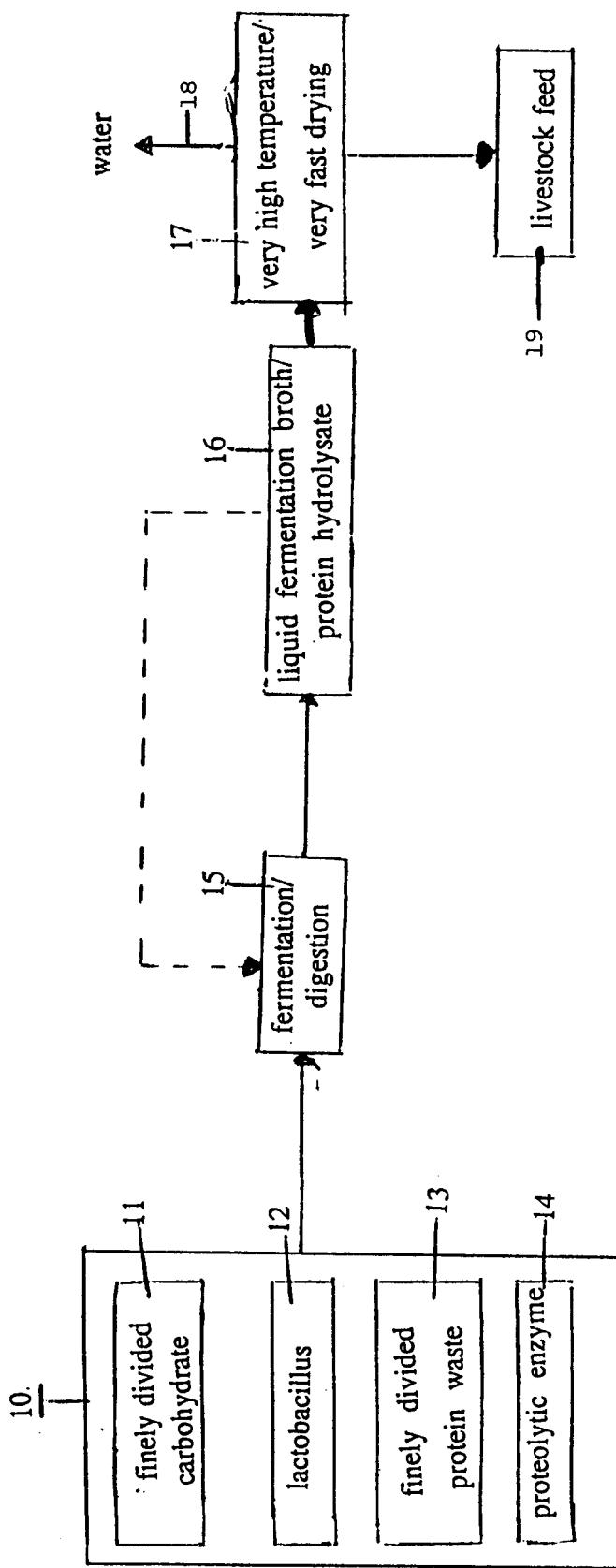


FIGURE 1

*Manure treatment wastes*

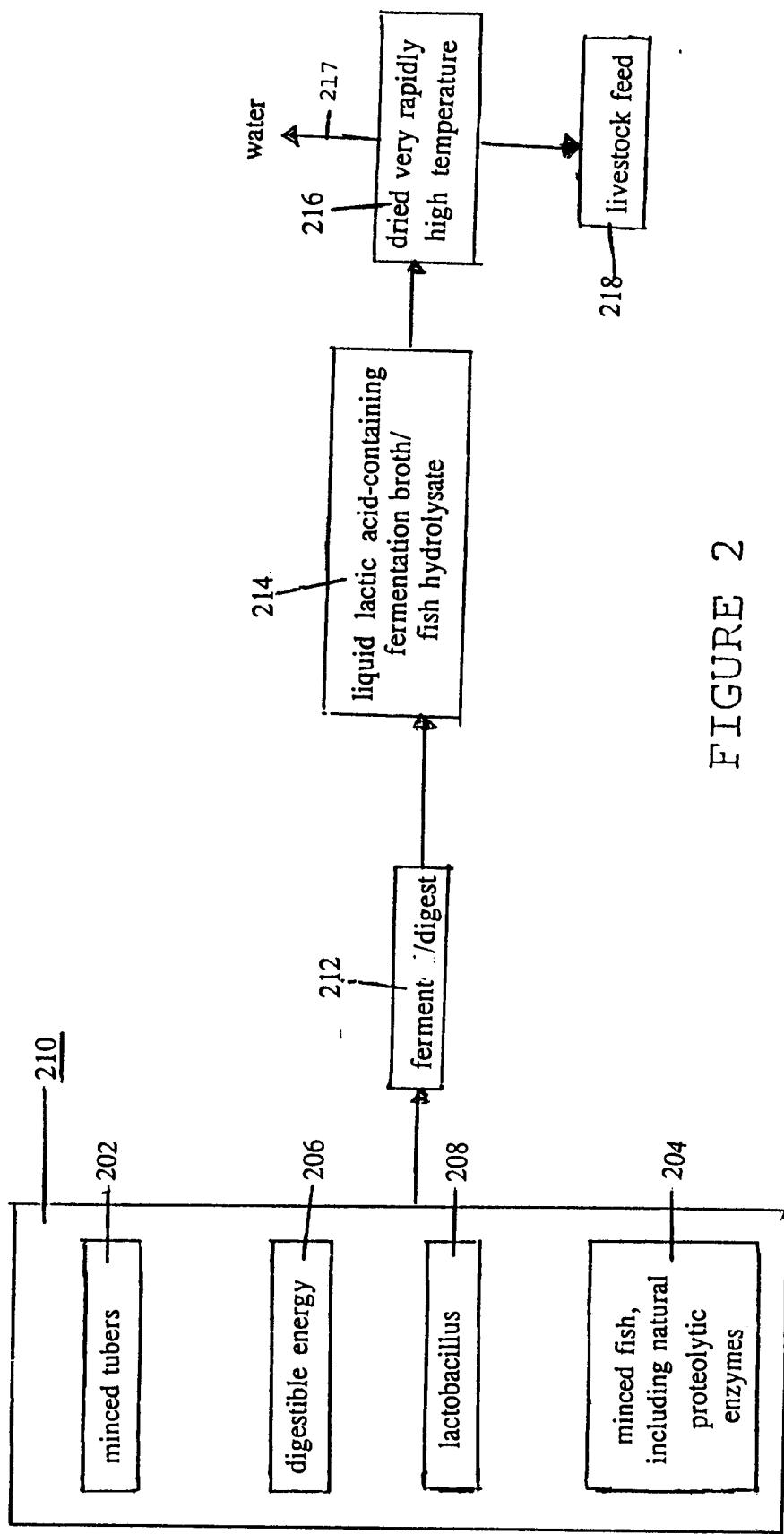


FIGURE 2

*Marcos + Horroates*

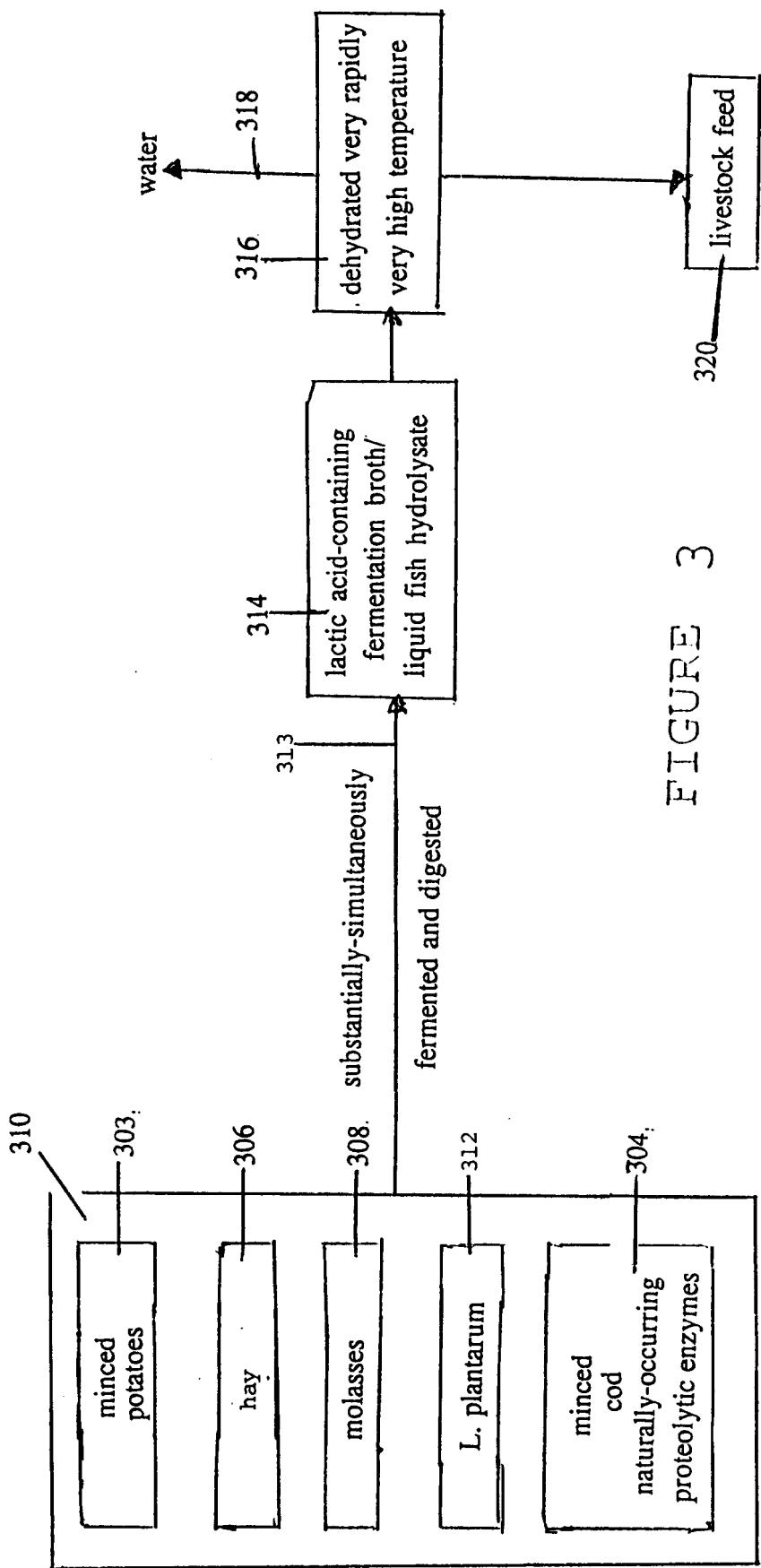


FIGURE 3

Marcos + Horrocastes

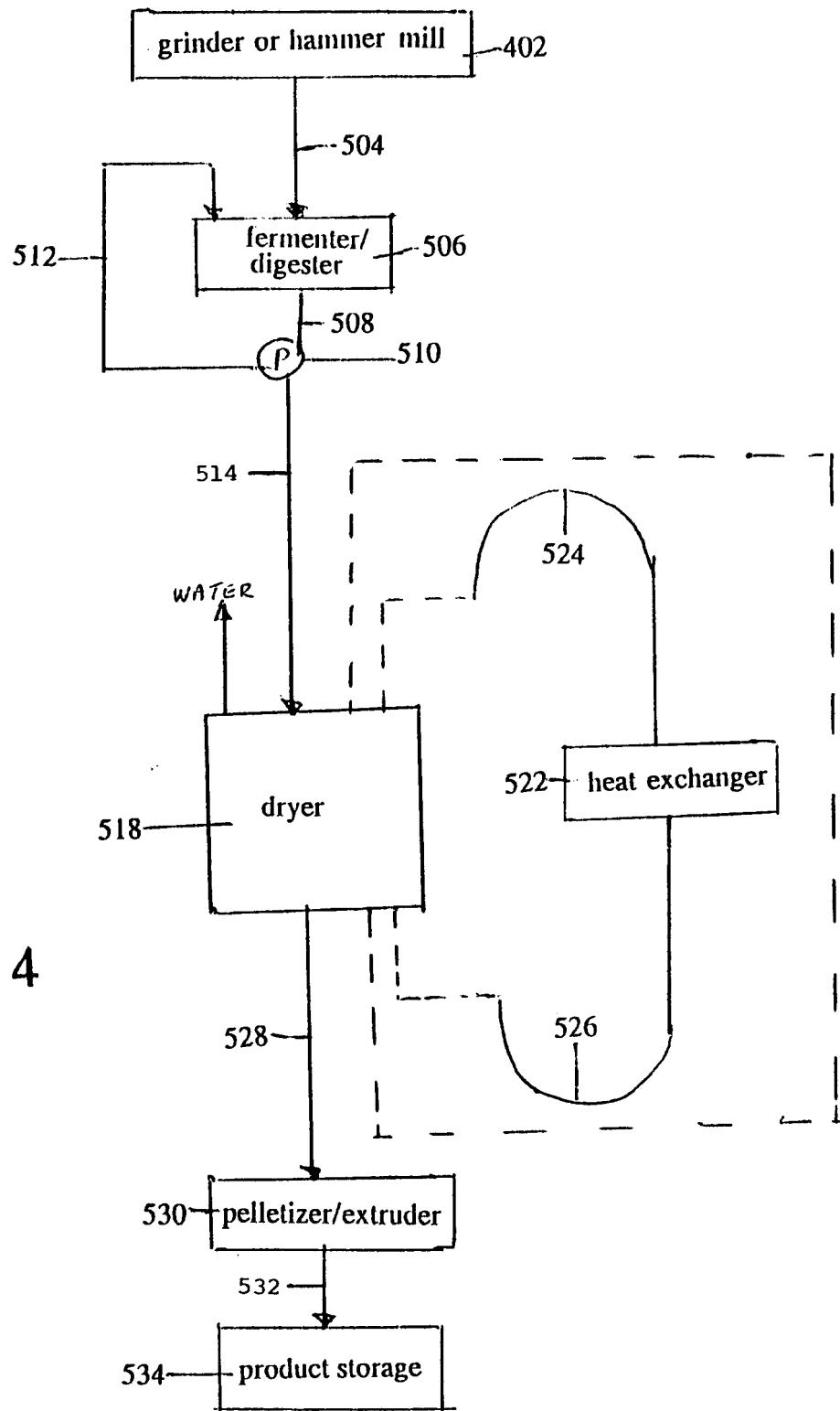


FIGURE 4

Marcus + Associates